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Nutrient Limitation in a Southwestern Desert Reservoir: Eutrophication of Las Vegas Bay, Lake Mead, Nevada

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ABSTRACT

Algal bioassay tests were conducted with *Selenastrum capricornutum* and natural algae on inner Las Vegas Bay, Lake Mead, Nevada, from December 1992 through September 1993, to identify any nutrient limitation in an area of the reservoir that has experienced problems associated with severe nutrient enrichment. Three areas were sampled based on a gradient of water quality conditions that existed in Las Vegas Bay (LVB). Disodium ethylenedinitrilotetraacetate (EDTA) significantly stimulated algal growth compared to non-EDTA treatment. Algal bioassays indicated that phosphorus (P) was the primary limiting nutrient at all stations for most of the test dates. Chl *a* response with EDTA + phosphorus (EDTA+P) was significantly greater ($p < 0.05$) than the control response. These tests suggest that continued P enrichment of lower LVB may well result in increased algal productivity.

INTRODUCTION

Lake Mead, Nevada, was formed in 1935 by impoundment of the Colorado River by Hoover Dam. Lake Mead has the largest surface area of any reservoir in the northern hemisphere and is used for flood control, irrigation, domestic and industrial water supply, power generation, navigation, recreation, fish and wildlife conservation, and sediment control (Hoffman and Jonez 1973).

The trophic status of lower Colorado river reservoirs ranges from oligotrophic to mesotrophic (Paulson and Baker 1984). Nutrient concentrations are low and phosphorus is often limiting. Nutrient fertilization experiments have been conducted to increase plankton productivity with the goal of improving the fisheries in sections of Lake Mead (Vaux and Paulson 1989). Much of Lake Mead is considered oligotrophic (Axler et al. 1987), although in the early 1970s eutrophication was thought to be in progress (Staker et al. 1974). U.S. Department of the Interior (1970) reported that Las Vegas Bay (LVB) was the most eutrophic section of Lake Mead. Today, LVB has been classified from hypereutrophic to oligotrophic based on chlorophyll and nutrient concentrations (LaBounty 1994).

Phosphorus deficiency in Lake Mead began to develop in 1963 when Glen Canyon Dam was constructed 460 km upstream. As the reservoir slowly filled, it substantially reduced the phosphorus supply in the Colorado River inflow to Lake Mead (Paulson and Baker 1980, Paulson et al. 1980, Evans and Paulson 1983). By contrast, lower sections of Las Vegas Bay, which lie within Boulder Basin have had severe problems with excess nutrient enrichment from point and non-point source pollution. Summer surface chl *a* concentrations often exceed 80 $\mu\text{g/L}$ (LaBounty 1994).

Eutrophication of LVB has progressed over the past 20 years. LVB is a collection bay for effluent from municipal- and county-operated secondary and advanced wastewater treatment plants discharged via Las Vegas Wash (LVW), a natural wash east of Las Vegas. Physical changes that have occurred in LVW have contributed to the increase of eutrophication in this area. In the early 1980s, floods eroded wetlands and drastically changed the morphometry of the wash (Roline and Sartoris 1988). Discharge in LVW increased from 2.9 m³/s in 1983 to 4.6 m³/s in 1992 and the temperature of the water entering LVB has increased (Sartoris and Roline 1993). Hydrodynamic interaction between LVW and LVB changed in the late 1980s from a cool water salinity plume via LVW, which at one time plunged below the euphotic or epilimnetic waters (Fischer & Smith 1983), to a less dense warm water salinity plume that now permeates the epilimnetic waters during the warmer months of the year. Under such circumstances, the nutrients contained in the LVW inflow are injected directly into the epilimnetic waters of LVB at the beginning of the growing season (French 1989).

Previous bioassays have been conducted on Lake Mead. Prentki and Paulson (1983) reported the upper basin of Lake Mead was phosphorus (P)-limited and the lower basin, LVB, was nitrogen (N)-limited. These results were reported prior to operation of the advanced water treatment plant in 1982, which removes phosphorus from the area's secondary effluent. Greene et al. (1986) conducted algal bioassays in LVB and found it to be primarily P-limited. Objectives of this investigation were to 1) determine if nutrient limitation in LVB has changed from the early 1980s to the present by conducting algal bioassays, and 2) assess the potential effect of increased eutrophication on the natural algal assemblage.

METHODS

Water samples were collected eight times from three stations in LVB, from December 1992 to September 1993 (Fig. 1). These three stations were selected based on the trophic gradient that exists among them. Station LV1 was 7 m or less in depth, LV5 was 23 m or less, and LV14 was about 100 m. LV14 was the control station, since it was situated 7 km from the mouth of LVW and was presumably not affected by the salinity plume. Samples were collected from 0 to 5 m with a composite sampler. Water was transferred into opaque, acid-washed, non-metallic, 19-L containers and transported by overnight express to our Denver laboratory. Water samples were stored in the dark at 4°C and assayed within 2 d of collection.

Bioassays were conducted in 500-mL Erlenmeyer flasks containing 200 mL of test water under constant light. *Selenastrum capricornutum* Printz was used only in December 1992 to evaluate the effects of a chelating agent and various levels of nutrients on algal growth response measured as chl *a* (Miller et al. 1978). Final concentration of *Selenastrum* in each replicate was 1500 cells/mL. One set of samples was spiked with 1.0 mg EDTA/L (disodium ethylenedinitrilotetraacetate); the other set was not spiked. Four concentrations of NaNO₃ or K₂HPO₄ were used as nutrient additions; they were: NaNO₃ 1.00 (level 1), 10.00 (level 2), 100.00 (level 3), 1000.00 (level 4) mg/L, and K₂HPO₄ 0.05

(level 1), 0.50 (level 2), 5.00 (level 3), 50.00 (level 4) mg/L. Controls included an EDTA spike or raw water without nutrient additions. Test flasks were cultured for 14 d under constant light ($90 \mu\text{E m}^{-2} \text{s}^{-1}$) at $24 \pm 0.5^\circ\text{C}$. Cultures were shaken thoroughly twice a day.

Chl *a* concentration was measured as an indicator of treatment response. Ten mL of water from each replicate of the *Selenastrum* bioassay test were filtered through Whatman GF/C glass fiber filters. Chlorophyll was extracted with 90% cold acetone and measured fluorometrically on a Turner Design Fluorometer Model 10-005R.

Natural algae was used as a spike in the bioassay tests beginning in January 1993. January bioassays were conducted similarly to the *Selenastrum* test, except that water samples were not filtered. From March through September 1993 all water samples were filtered. Five liters of unfiltered water containing the natural algae from each station were used as spikes for the bioassays. These five liters were passed through a 26- μm plankton net; carbonated water was poured over the filtered plankton to asphyxiate the zooplankton. Large zooplankters were separated out from the phytoplankton. Phytoplankton were then washed with distilled water and were returned to the original five liters of water. Water was mixed thoroughly as 200-mL aliquots were dispensed into acid-washed 500-mL Erlenmeyer flasks. Triplicates were used for all levels of treatment and inoculated with 5 mL of the natural algae. Number of cells per mL varied with sampling date. Lovstad (1983) used this technique to enable the cells to grow exponentially for at least 3 to 5 d without affecting the composition of the medium. Bioassays were conducted under a light:dark cycle of 12:12 or 13:11 h.

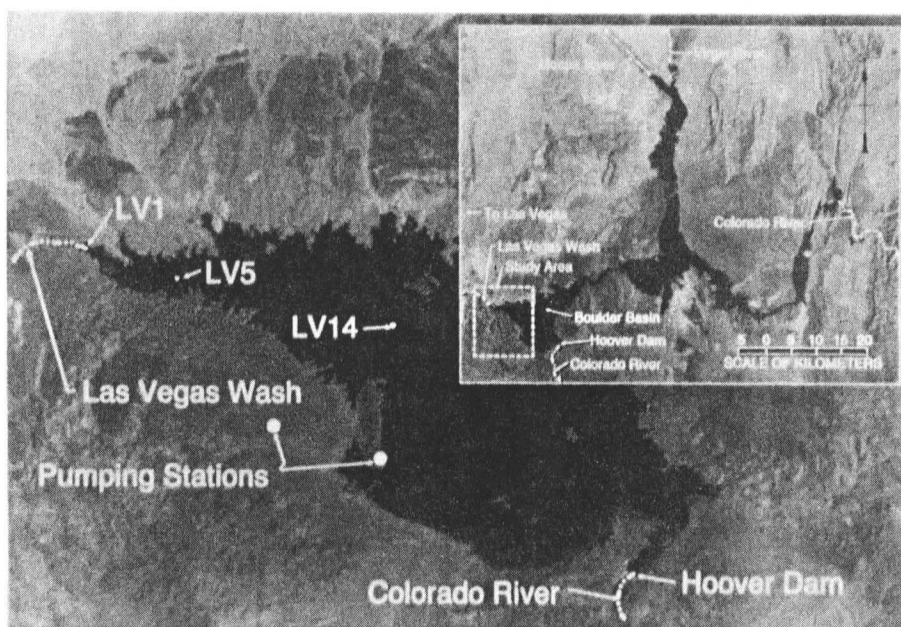


Figure 1. Location map of Lake Mead, Nevada showing the study area, Las Vegas Bay, within Boulder Basin. Las Vegas Wash flows into LVB, where the test waters were sampled from three stations, LV1, LV5 and LV14.

Nutrient treatment levels identical to the one described for the *Selenastrum* test were used for the natural algal bioassays from January through April 1993. Bioassays conducted from May through September 1993 were spiked with nutrients at levels 1 and 2 only; treatment level replicates were increased from three to six. The initial algal bioassays demonstrated that the two lowest levels of treatment resulted in increased growth response and the highest treatment level inhibited algal growth. EDTA and non-EDTA controls were set up without nutrient enrichment spikes in January 1993. Thereafter, all controls were spiked with EDTA from March through September 1993. Temperatures in the growth chambers were maintained at ambient reservoir temperatures that ranged from 17 to 28 °C, and light intensity was about 255 $\mu\text{E m}^{-2} \text{s}^{-1}$. Thirty mL from each replicate were filtered through Whatman GF/C filters for chl *a* analysis. All replicates were sampled at least three times over the 2 to 13 d period.

Water samples were routinely analyzed for chl *a* (Strickland and Parsons 1968) and nutrient concentrations (U.S.Environmental Protection Agency 1979) from a composite stratum of 0 to 5 m at the three reservoir stations.

Chl *a* concentrations from bioassay tests were log transformed. Dunnett's one tailed t-test was used in analysis of bioassays (SAS Institute 1987). The statistical approach was to test a number of treatment means versus a single (common) control in which each treatment represented the addition of a particular nutrient at a specified level of concentration. Thus, multiple comparisons were made - each treatment mean versus the single control. A one-tailed test was used because detecting an increase in chlorophyll productivity was desired. Significance was determined at the $p < 0.05$ level.

RESULTS

Selenastrum vs. natural algal bioassays

The addition of level 2 EDTA+P to the *Selenastrum* bioassays increased the chl *a* response significantly ($p < 0.05$) compared to those samples that were not spiked with the metal chelator (Table 1). Results indicated that waters were metal laden. *Selenastrum* and natural algal bioassays were run a month apart, which may have accounted for the significant increase ($p < 0.05$) in the chl *a* response with natural algae, although chl *a* concentration measured at LV1 showed a decrease from 1.25 $\mu\text{g/L}$ (Dec.) to 0.34 $\mu\text{g/L}$ (Jan.).

Table 1. Comparison of chlorophyll *a* ($\mu\text{g/L}$) response with EDTA versus non-EDTA treatment and level 2 nutrient enrichment. C = control. PEDTA = P + EDTA. NEDTA = N + EDTA.

Spike	Station	EDTA	C	PEDTA ^a	P	NEDTA	N
<i>Selenastrum</i>	LV1	0.62	0.19	20.61	1.20	0.25	0.16
	LV5	0.28	0.05	15.99	4.18	0.29	0.04
	LV14	0.04	0.04	14.81	1.09	0.04	0.04
Natural algae	LV1	2.65	0.34	43.56	3.85	12.51	1.25
	LV5	0.29	0.16	44.29	0.22	0.11	0.08
	LV14	0.42	0.05	14.50	0.66	0.93	0.07

^a chl *a* response with PEDTA was significantly greater ($p < 0.05$) than C for *Selenastrum* and natural algal bioassays at all stations.

Maximum chl *a* response for *Selenastrum* occurred at day 7 for LV1 and LV5, and day 14 for LV14 (Fig. 2). A significantly greater maximum response ($p < 0.05$) occurred in levels 1, 2, and 3 EDTA+P treatment compared with the control at all stations. Level 4 EDTA+P appeared to inhibit a response from both the *Selenastrum* and natural algal bioassays.

Station LV1

Maximum chl *a* response with level 2 EDTA+P treatment was significantly greater ($p < 0.05$) than the response with the control, which indicates P-limiting conditions for the March bioassay test (Fig. 3). N and P concentrations were high in the upper 5 m of the water column, particularly NH_3 and $\text{PO}_4\text{-P}$. The ratio of TIN (9.53 mg/L): $\text{PO}_4\text{-P}$ (1.20 mg/L) for March was 8:1, which indicated the waters were N-limiting. Although data indicated N-limiting conditions, chl *a* response with EDTA+nitrogen (EDTA+N) treatment did not reflect these results. The April bioassay test indicated that P was the primary limiting nutrient. Chl *a* response with level 1 EDTA+N treatment exceeded the control significantly on day 6, although the primary limiting nutrient for the April bioassay was EDTA+P. The sample appeared to be N-limiting initially and then with growth became primarily P-limiting. Level 2 EDTA+P treatment stimulated a significant maximum response compared with the control on day 6, which indicated P-limiting conditions for May (Fig. 3). Chlorophyll response to level 1 and 2 EDTA+P treatment for the June and July bioassay was significant over the control (Fig. 3). EDTA+P bioassay response greatly exceeded the measured composite chl *a* concentration of 41 $\mu\text{g/L}$ recorded in July from station LV1. In August, the chl *a* response to level 1 and 2 EDTA+P treatment was significantly greater than the control (Fig. 3). These responses indicated P was the primary limiting nutrient. Levels 1 and 2 EDTA+P treatment were not able to sustain a response after day 4, which demonstrated nutrients may have been depleted by then. In September, levels 1 and 2 EDTA+P treatment stimulated a significantly greater response ($p < 0.05$) compared to the control from days 2 to 5; EDTA+N treatment and the un-enriched control also were stimulated (Fig. 3). The September bioassay test was a good example of rapid growth followed by decline that occurred for most of the bioassay tests conducted during the study period.

Station LV5

Level 2 EDTA+P treatment stimulated a significant chl *a* response ($p < 0.05$), reaching a maximum of 28 $\mu\text{g/L}$ in March (Fig. 3). By contrast, the bioassay test at LV1 showed tremendous growth with EDTA+P due to the mixing of nutrients throughout the water column. In April, no chl *a* response occurred with N or P treatment. This lack of response may indicate that the water was neither N nor P limited, even though water samples collected from LV5 showed the ratio of TIN (2.27 mg/L): $\text{PO}_4\text{-P}$ (0.03 mg/L) was 75:1, indicating P limiting conditions. Bioassay tests in May demonstrated that chl *a* responded significantly to level 1 EDTA+P treatment compared to the control. For the June test, level 2 EDTA+P treatment resulted in a growth response to 270 $\mu\text{g/L}$ chl *a*, followed by a rapid decline. This response was the highest for the entire sampling period and far

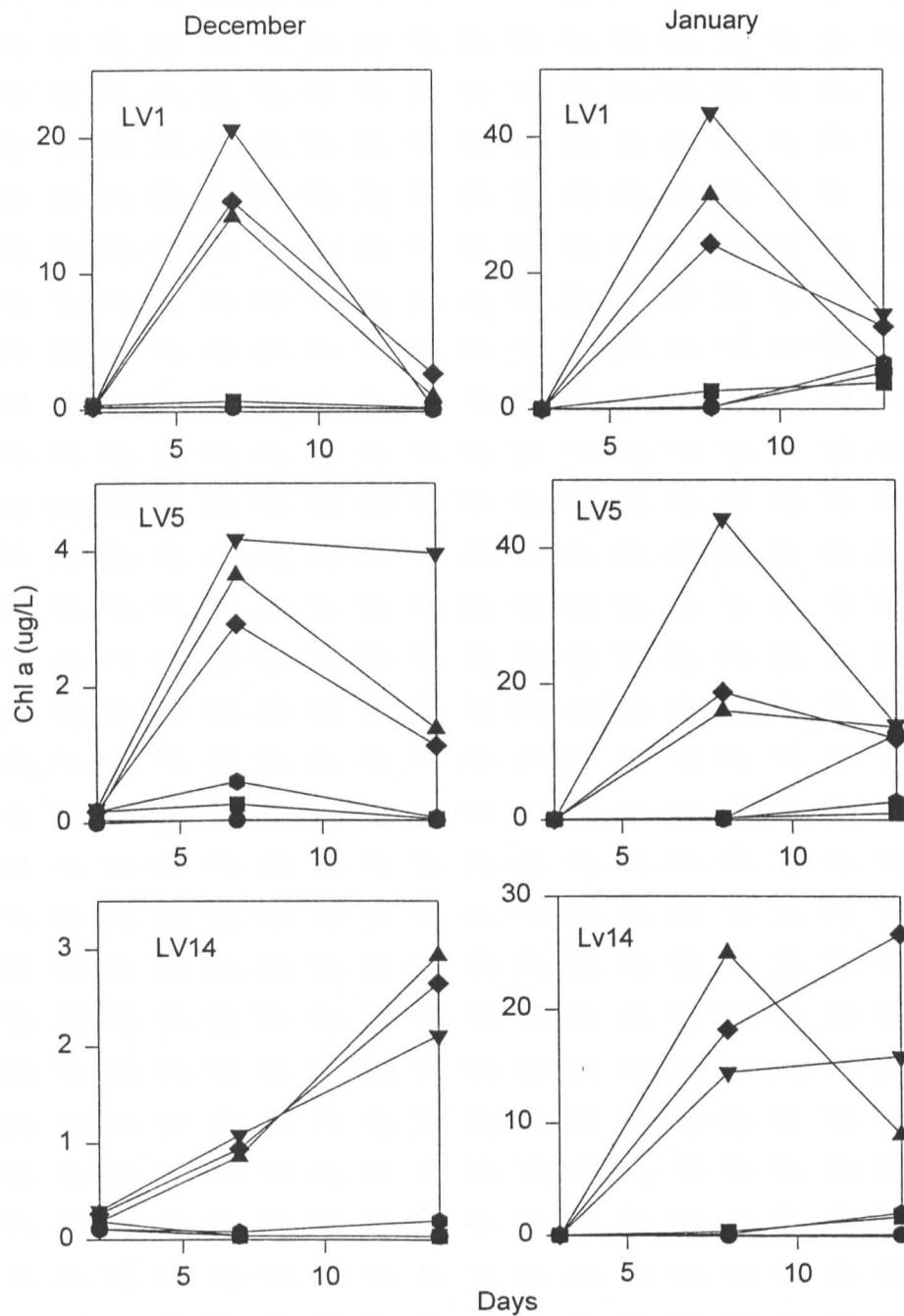


Figure 2. Comparison of levels 1,2,3 and 4 EDTA+P treatment versus EDTA+control on the chl a response with test waters from stations LV1, LV5, and LV14. *Selenastrum* bioassay test was conducted in December 1992. Natural algae was used from each respective station in the January 1993 bioassay test. Control (○); EDTA+control (■); EDTA+P: level 1 (▲), level 2 (▼), level 3 (◆), level 4 (●).

exceeded the measured chl *a* concentration of 18 µg/L in the reservoir. August maximum response with level 2 EDTA+P treatment was significantly lower ($p < 0.05$) than the chl *a* responses from June and July. Both levels of EDTA+P treatment resulted in significantly higher chl *a* concentrations ($p < 0.05$) compared to the control for the September bioassay. Response was well above the 18 µg/L chl *a* concentration that was measured at LV5. Algal bioassays from LV5 indicated P-limited conditions for most of the test period.

Station LV14

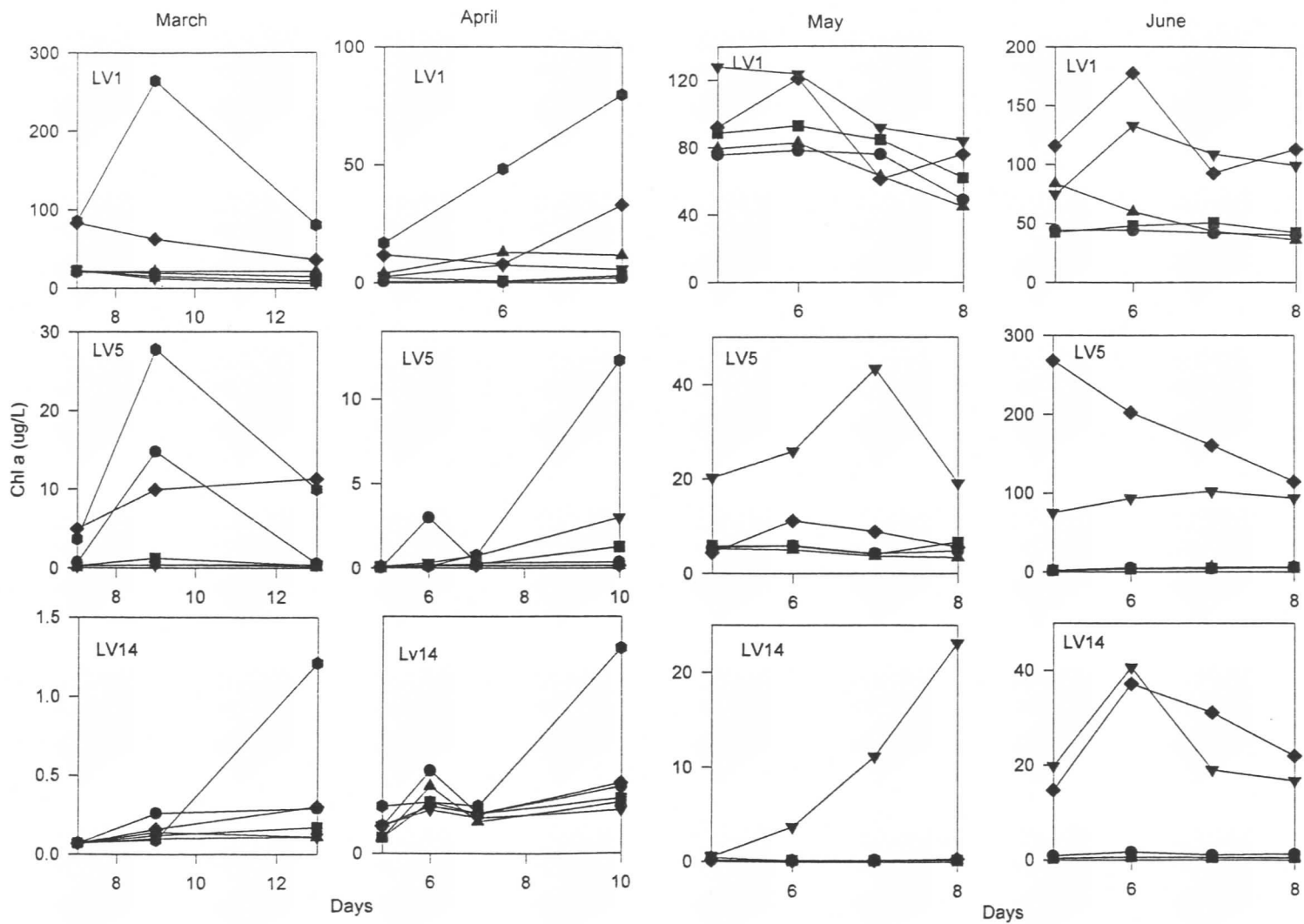
Maximum chl *a* response generally did not exceed the chl *a* responses reached at the other two stations (Fig. 3). Greatest maximum chl *a* response (75 µg/L) for the sampling period was attained in July for level 2 EDTA+P treatment. In contrast, the maximum chl *a* response (270 µg/L) was reached in June at LV5, and in March at LV1 (270 µg/L). Chlorophyll *a* concentration measured at station LV14 was generally under 2 µg/L for the entire sampling period.

DISCUSSION

Selenastrum bioassays indicated that EDTA was required to reach maximum growth potential when nutrients were limiting. Greene et al. (1986) conducted nutrient enrichment tests with *Selenastrum* on water samples collected from Lake Mead in 1979 and found that concentrations of Ni, Zn, Cu, and Pb were high enough to cause toxicity. Greene et al. (1986) stated that because natural algae is adapted to the metals, indigenous algal growth was probably controlled by nutrient limitations rather than by heavy metal inhibition. In this study, the *Selenastrum* and natural algal tests showed that the chl *a* response was significantly greater with EDTA+P treatment over the control at all three stations. Rapid decline in chl *a* after maximum response was achieved may have been caused by the P-limiting conditions that reoccurred in the samples after the initial spike. Aldridge et al. (1993) stated that other processes associated with nutrient deficiency stress include actual loss of phytoplankton biomass from cell death or chlorophyll pigment loss due to chlorosis.

Productivity in LVB was largely influenced by the interaction between LVW and LVB. Exchange between LVW and LVB and the influence on epilimnetic waters was important in determining algal productivity at each station. The ratio of $\text{TIN}:\text{PO}_4\text{-P}$ averaged 24:1 at LV1 during the sampling year. A $\text{TIN}:\text{PO}_4\text{-P}$ ratio of greater than 14 indicates a largely phosphorus-limited environment: a ratio of less than 10 indicates a nitrogen-limited environment (Lambou et al. 1976).

Station LV1 was influenced by LVW more than the other two stations because of the constant mixing of the water column; this influence was demonstrated in the algal bioassays. The salinity plume generally entered LVB via LVW at the bottom of LV1 due to the shallow depth of the station, lack of thermocline, and warmer water temperatures in LVB than in LVW. The salinity plume gradually rose to immediately below the surface in June and July 1993. Nutrient loads entering LVB via LVW were 110 kg/d of $\text{PO}_4\text{-P}$, 4800 kg/d of $\text{NH}_4 + \text{NH}_3$, and 1100 kg/d of $\text{NO}_3\text{-N}$ (Sartoris and Roline 1993). Nutrient concentrations were higher in July than in other months. The salinity plume plunged to the bottom depths of LV5 from October through February and then was detected above the



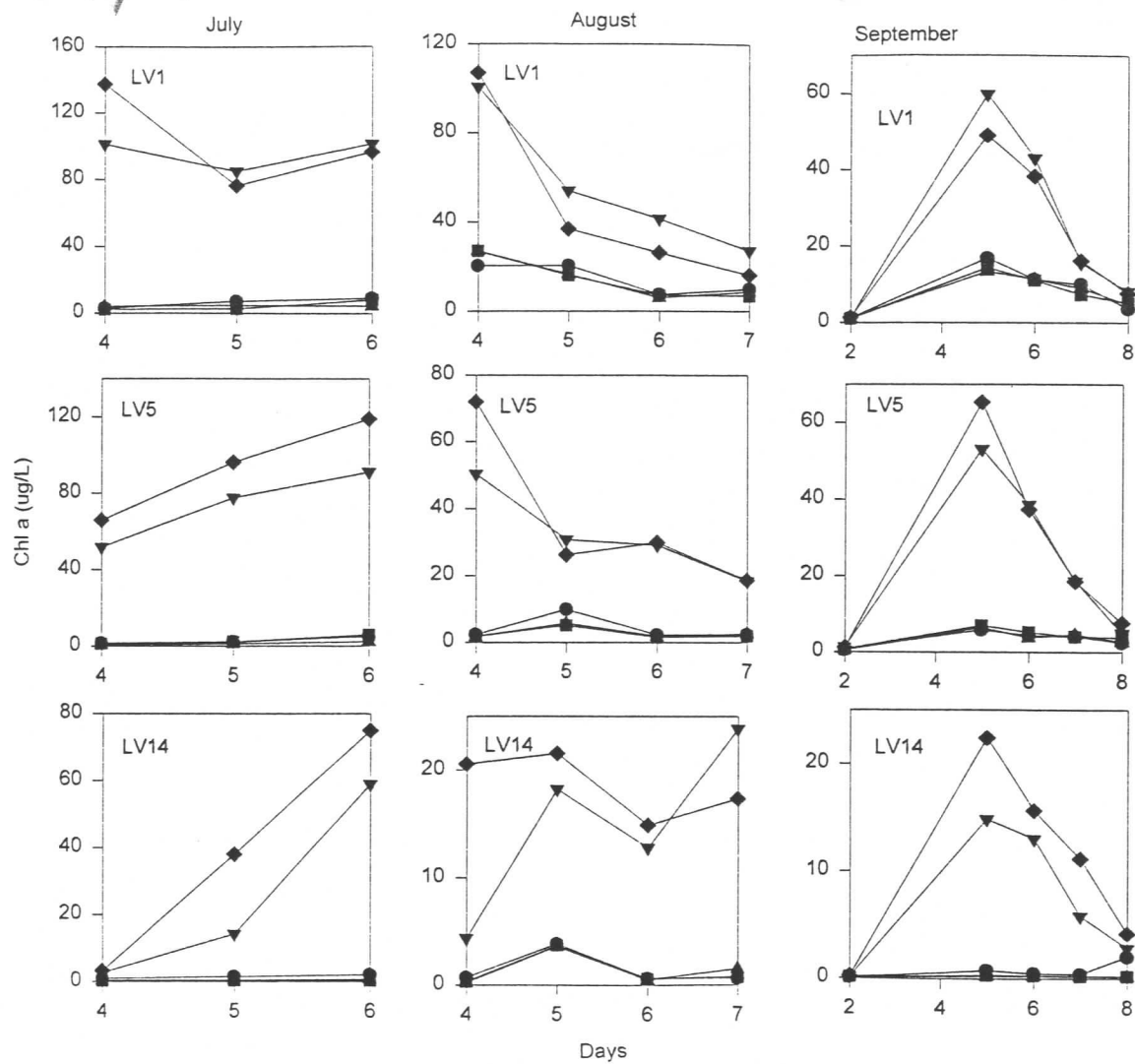


Figure 3. The response of natural algae chl *a* with levels 1 and 2 EDTA+P and EDTA+N treatment versus EDTA+control on test waters from March through September 1993.

thermocline at 9 to 13 m during March through September, which allowed nutrients to mix in epilimnetic waters. $\text{PO}_4\text{-P}$ and TIN were less concentrated at LV5 than at LV1 because of an increased water volume. The TIN: $\text{PO}_4\text{-P}$ ratio averaged 61:1 at LV5. The salinity plume did not appear to affect station LV14 directly; productivity was low compared with the other two stations, and was a good indicator of nutrient limitation throughout Lake Mead. N to P ratios at all three stations indicated P-deficiency conditions, due mainly to the high concentration of TIN and low concentration of $\text{PO}_4\text{-P}$ at all of the stations.

Station LV1 was highly productive. The addition of P to the bioassays from station LV1 resulted in greatly increased production. Maximum responses from EDTA+P treatment were almost always greater than the measured chl *a* concentration at station LV1. For six of the test dates, the maximum chl *a* response was higher at LV1 than at the other two stations. Why did limiting conditions exist at LV1 when the P concentrations were already high and the chl *a* concentrations indicated hypereutrophic conditions? Why did additional P enrichment cause further growth in chlorophyll? One explanation may be that P entering the inner bay plunged below the depth of maximum algae and was not directly available for assimilation by the algae. This explanation is not likely because both the water samples for the bioassays and the nutrient analyses were composite samples and therefore reflected the nutrient concentration in the water column, particularly at LV1, where summertime depths were usually less than 2 m. If P governs the algal production in inner LVB, then increased P concentrations would mean greater chl *a* concentrations and increased hypereutrophy. The effect of increased primary production on the striped bass fishery in inner LVB would be substantial (Mueller 1993), but increased eutrophication would result in degraded water quality of the inner bay (LaBounty 1994).

The number of year-round algal bioassay tests conducted was not sufficient to allow a conclusive statement about seasonal nutrient limitation at station LV1. A chl *a* response to the EDTA+N treatment for the March, April, May, June, and September bioassay tests may indicate N was a secondary limiting nutrient. April and June were the only months in which the EDTA+N treatment caused a significant response ($p < 0.05$) in chl *a* compared to the control. The only two months that EDTA+N did not exceed control chl *a* response were July and August. Algal bioassays indicated that LV1 was P limiting for most of the sampling dates.

In only June and September did the maximum chl *a* response at LV5 exceed LV1. The greater depth of station LV5 may have diluted nutrient concentrations in the water column. Maximum chl *a* response at LV5 was less than that measured for LV1, but was sustained for a longer period and did not decline as rapidly as did the responses at LV1. Station LV5 has been classified as hypereutrophic (LaBounty 1994). This station is a good example of how the salinity plume travels from LVW into LVB, depending on the depth of the thermocline, and influences the primary productivity. As the waters began to stratify in March 1993 the salinity plume was tracked by the specific conductance

profile (LaBounty 1994, pers. comm.). During the warmer months the salinity plume reacted as a neutral or buoyant jet guided by the density of the water. When the nutrient-laden jet of LVW water entered the bay above the thermocline, the epilimnetic waters were enriched with nutrients. The extent of nutrient enrichment depended on the depth of the epilimnion. When the salinity plume entered below the thermocline, nutrients were not available to the algae, and productivity was generally reduced. For the duration of the study, the salinity plume often mixed with the epilimnetic waters. During March, the salinity plume was observed at 13 to 15 m, which created an ectogenic meromixis condition caused by saline water entering freshwater. The inflowing salinity plume resulted in higher temperatures in the water column at the point where the salinity plume was observed. This condition was observed only during the March sampling period. Nutrients from 0 to 5 m depth ($\text{PO}_4\text{-P} = 0.10$, $\text{NO}_3\text{-N} = 1.73$, $\text{NH}_3\text{-N} = 0.53$) were at lower concentrations than the nutrients in the sample collected in the salinity plume at 14.5 m ($\text{PO}_4\text{-P} = 0.10$ mg/L, $\text{NO}_3\text{-N} = 3.10$, $\text{NH}_3\text{-N} = 2.80$). The ratio of $\text{TIN}:\text{PO}_4\text{-P}$ was 75:1 for 0 to 5 m and 59:1 for 14.5 m. This ratio illustrates how a greater concentration of nutrients occurred below the thermocline as a result of the density difference of the inflowing salinity plume.

LV14 was considerably less productive than the other two stations when comparing phytoplankton, chlorophyll, and nutrient concentrations that have been collected from Lake Mead (LaBounty 1994, pers. comm.). Station LV14 was oligotrophic and indicative of the trophic status of most of Lake Mead. At all stations, algal production was P-limited from May through September.

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